Changes in biomass, chlorophyll content and gas exchange of beans and peppers under nitrogen and water stress

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Abstract

We experimentally produced moderate water stress that reduced leaf, stem, root and fruit biomass of peppers, and severe nitrogen (N) limitations that almost stopped their development. Root/shoot ratios (R/S) were higher in N-limited plants. Low water availability (and also low N availability) produced lower stomatal conductance (g_s). Specific leaf mass was higher and chlorophyll (Chl) concentration was lower under low N-availability. The same experimental conditions produced smaller differences among treatments in beans (with N-fixing symbionts). Water stress increased its relative importance as shown by the induced increase in root/shoot ratio. N stress was less important as shown by the absence of effects on Chl concentrations and g_s. Both peppers and beans responded to limited availability of nitrogen and water by allocating to structures involved in uptake (roots), by longer organ duration, and by increasing the efficiency of N and water use.

Introduction

The influence of both nitrogen and water deficiencies in plant production is well documented (Hanks and Rasmussen 1982), but the effects on basic processes are less known. In crop plants experiencing N and water stress, alterations occur in the distribution of assimilates between shoot and roots. The effect most often found is an increase in the partitioning of saccharides to roots, implying decreases in shoot to root growth ratios (Brouwer 1962, Wilson 1988). Partitioning of the above-ground parts also varies; thus changes in specific leaf mass are common (McDonald 1990). Chl content and g_s are also decreased. Therefore, smaller growth and yield are the final result (van Keulen et al. 1989).

Fertilization and irrigation experiments show that, up to a certain level, addition of N fertiliser and watering leads to higher yields. But, interpretation of these experiments seldom exceeds the derivation of the optimum application rate for the

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particular conditions of the experiment. There is a lack of explanatory and predictive conclusions.

Our aim was to study the changes in biomass accumulation and partitioning in N and water stressed plants in order to obtain new explanatory insights for horticultural plants. We compared nitrogen-fixing plants, beans, with non-nitrogen-fixing plants, peppers, to study different biological responses to the same fertilization and irrigation treatments. We focused on responses tending to ameliorate the limitation by specifically targeting the limiting resource such as allocation to the structures involved in uptake, the organ duration, and the efficiency with which nitrogen and water are used (Chapin et al. 1987).

Materials and methods

Seeds of *Phaseolus vulgaris* L. cv. Garrofal and *Capsicum annuum* L. pure line B6 were grown in controlled environment chambers. Plants were grown in a chamber on a 14/10 h light (300 µmol m⁻² s⁻¹)/dark cycle, 70 % relative humidity and 25/20 °C day/night during four months. Plants were grown in 2 500 cm³ containers in a substrate consisting of 1:1 (v:v) peat moss and sand. A 2² factorial experiment was carried out by establishing two levels of irrigation and two levels of nitrogen supply. Water-stressed plants were fertirrigated only when soil matric potential reached -0.04 MPa ("low water" treatment - LW) and control plants when it reached -0.01 MPa ("high water" treatment - HW). The two levels of N supply were established by irrigating with "high nitrogen" solution (HN) containing [keq m⁻³]: 4.88 N0₃⁻, 0.63 H₂PO₄⁻, 4.41 SO₄²⁻, 2.76 K⁺, 8.71 Ca²⁺, 2.01 Mg²⁺, 1.36 NH₄⁺, 17.8 Fe²⁺, 9.09 Mn, 1.82 Zn, 7.87 Cu, 4.62 B, 1.04 Mo, 25 Mg. The low nitrogen (LN) solution had only 1.2 keq m⁻³ NO₃⁻ and the same concentrations of the other elements. Growth continued for 3-4 months.

The height of plants and the number and area of leaves were measured every week. Other data for plant growth analysis were obtained from two harvests (43 and 122 d after germination in peppers and 29 and 98 d after germination in beans). In each harvest, fresh and dry mass of different organs (roots, stems, leaves and fruits) of 4 plants from each treatment were measured. Leaf area of these plants was measured with a leaf area meter *LI-COR 3000* (LI-COR, Lincoln, NE, U.S.A.). Plant components were then dried at 60 °C in an oven until constant mass was reached. Specific leaf mass (SLM) was also determined.

Transpiration rates (E), gₛ, and leaf temperatures were measured with a *LI-COR* model *LI-1600* steady-state porometer in 6 top canopy well developed leaves of each treatment every 3 or 4 d.

A *SPAD* chlorophyll meter from *Minolta* (model 502) was used to measure leaf Chl concentrations (Monje and Bugbee 1992). The relative units were previously calibrated measuring leaf Chl concentrations by shaking fresh leaf discs in N,N'-dimethylformamide (Porra et al. 1989).
Results

Our experimental design produced severe nitrogen limitations in LN treatments and moderate water limitations in LW treatments. Plant growth and assimilates partitioning changed to compensate the N and water stresses.

Stressed plants had a lower biomass in all their organs (roots, stems, leaves, and fruits) (Fig. 1). The decrease in biomass produced by N limitation was more expressed than the decrease produced by water limitation in both species, but the difference was relatively smaller in beans. LN plants accumulated much less biomass than HN plants (more than 5 times lower in peppers, and about 3 times lower in beans at the end of the growth cycle). In fact, peppers (without N-fixing symbionts) suffered severe N stress that stopped almost completely the development (Fig. 1).

![Graph showing biomass distribution of Phaseolus and Capsicum](image)

**Fig. 1.** Total, leaf, stem, root and fruit dry masses in beans and peppers exposed to different fertirrigation treatments. Data correspond to two different harvests throughout the growth cycle. DAG = days after germination. LW - Low Water, HW - High Water, LN - Low Nitrogen, HN - High Nitrogen (for description of treatments see Materials and methods). SE bars are also represented.

The unexpected large decrease in beans was only found at the end of the growth cycle and it was mainly due to lower stem dry mass, and to a decay of root symbiotic nodules that decreased in number and function. There was also important leaf loss...
during flowering and fruiting as shown by the lower leaf area in the second harvest.

In N-limited peppers, root biomass decreased relatively less than the above-ground biomass. Therefore, they showed higher root/shoot (R/S) ratios (Fig. 2). Similar trends were found for water-stressed beans (Fig. 2). SLM was higher and Chl concentration lower in N-stressed peppers but they were not lower in N-stressed beans (Fig. 2).

![Graph showing root/shoot ratio, leaf area, specific leaf mass, chlorophyll (Chl) concentration, and stomatal conductance (g_s) in beans and peppers exposed to different fertirrigation treatments. Data correspond to two different harvests throughout the growth cycle. For explanation of symbols see Fig. 1.]

Lower water availability produced lower g_s and higher leaf temperatures (0.69 ± 0.10 °C higher in LW treatment versus 0.08 ± 0.10 in HW treatment). Similar effects, although weaker, were produced by low N availability (Fig. 2). Limited availability of N and water required for plant growth elicited responses that tended to ameliorate the limitation. Some of the mechanisms specifically targeted the limiting resources. There was an increased relative allocation to the structures involved in uptake such as roots; the ratio root/shoot decreased with increasing N supply (Fig. 3, top). There was an increased organ duration to decrease the rate of resource loss: stressed plants had less leaves and these leaves grew less and lasted longer (Fig. 4). We used total dry mass over water supply as water use efficiency (WUE) and total dry mass over N supply as nitrogen use efficiency (NUE). Thus, we found that NUE was reduced by water stress, as shown by the lower NUE of LW treatments; and we found that NUE
Fig. 3. Root/shoot ratio (top) and total dry mass (bottom) versus nitrogen and water supply in beans and peppers exposed to different fertirrigation treatments. DAG = days after germination. SE bars are also represented.
was increased by N stress, as shown by the higher NUE of peppers *versus* beans, even though data corresponded to the second harvest when beans were old and had lower N-fixation than in the first periods of the growth cycle (Fig. 3, *bottom*). NUE was higher in peppers than in beans in consonance with their higher N-limitation. WUE was decreased by N stress, as shown by the lower WUE of LN treatments (Fig. 3, *bottom*).

![Graph showing average leaf area and leaf number over time for Phaseolus and Capsicum](image)

*Fig. 4. Leaf number and leaf area in beans and peppers exposed to different fertirrigation treatments throughout the growth cycle. For explanation of symbols see Fig. 1.*

**Discussion**

Plants responded to nutrition and hydric stresses by clearly decreasing productivity and by changing assimilation and partitioning of biomass and resources. The N limitation produced more severe effects on peppers than on beans in consonance with the absence of N-fixing symbionts. N-stressed peppers even stopped their development.

In N-limited plants (and also in water-limited plants, although not as much) growth slowed and was finally smaller, thus decreasing the sink strength (MacAdam *et al.* 1989). Our results are in apparent agreement with the concepts advanced by Brouwer (1963) and Raper *et al.* (1978) who viewed shoot and root growth in terms of functional balance controlling a plant’s supply of C and N. Plants that grow supply N to meristematic regions. Under low N availabilities, transport of N to shoot meristematic centres decreases, along with the availability of growth regulators.
(Kuiper 1988). As a result, meristematic activity and initiation of new leaves decline. This might have been the reason why our LN peppers did not grow (Fig. 4). Furthermore, growth and saccharide metabolism of existing sink tissues in the shoot decline. With decreased demand of saccharides in the shoot, a larger proportion of available photosynthate is translocated to roots. Thus, in our experiments partitioning between leaves and other above-ground organs also changed under N deficiency as has often been found by other authors (Boatwright and Haas 1961, Campbell et al. 1983). Leaf mass ratio decreased 33 % in peppers and only 15 % in beans (second harvest).

Nitrogen deficiency produced a smaller increase of $g_s$ in consonance with the results of Radin and Ackerson (1981) and Radin (1983) who found stomatal closure at much higher plant water potentials in N-deficient plants than in plants adequately N-supplied. Lower $E$ due to N deficiency may either be caused by stomatal closure at higher leaf water potentials or by lower root conductivity (Van Keulen et al. 1989). In any case, there appears to be a linear relationship between $g_s$ and N concentrations in the leaf (Yoshida and Coronel 1976).

LW plants seemed to avoid the stress by reducing water losses. They presented lower $g_s$, and consequently higher leaf temperatures (Peñuelas et al. 1992). However, the water stress we created in our experiments was moderate as shown by the minimum leaf water potentials we measured (-1.5 MPa of $\Psi_w$). This experimental water stress, although milder than the N stress, also produced severe decreases of growth and yield, likely because the cells decreased their turgor (data not shown) and that affected the rate of cell expansion and ultimately the cell size, the irradiated surface, and the photosynthates produced.

Both stresses, the one created by low N availability and the other by water deficiency, produced decreases in $g_s$, reallocation of biomass and decreased growth and yield.

Limited availability of N and water required for plant growth elicited responses that tended to ameliorate the limitation. Some of the mechanisms specifically targeted the limiting resources (Chapin et al. 1987). There was increased allocation to the structures involved in uptake such as roots (Fig. 3, top), increased organ duration to decrease the rate of resource loss (Fig. 4), and increased efficiency with which nitrogen and water were used (Fig. 3, bottom). Maintaining or increasing the consumption of nonlimiting resources in order to increase the efficiency with which a limiting resource is used is one of the mechanisms that horticultural plants used to resource-rich environments might use to maintain growth in resource-poor conditions.

References


